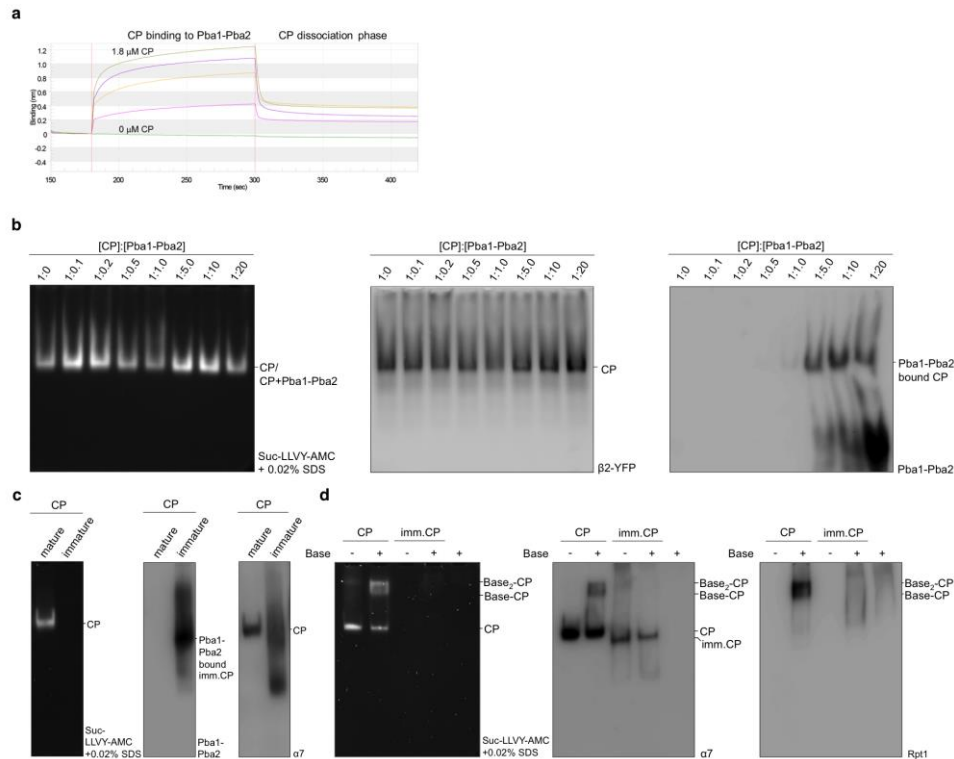


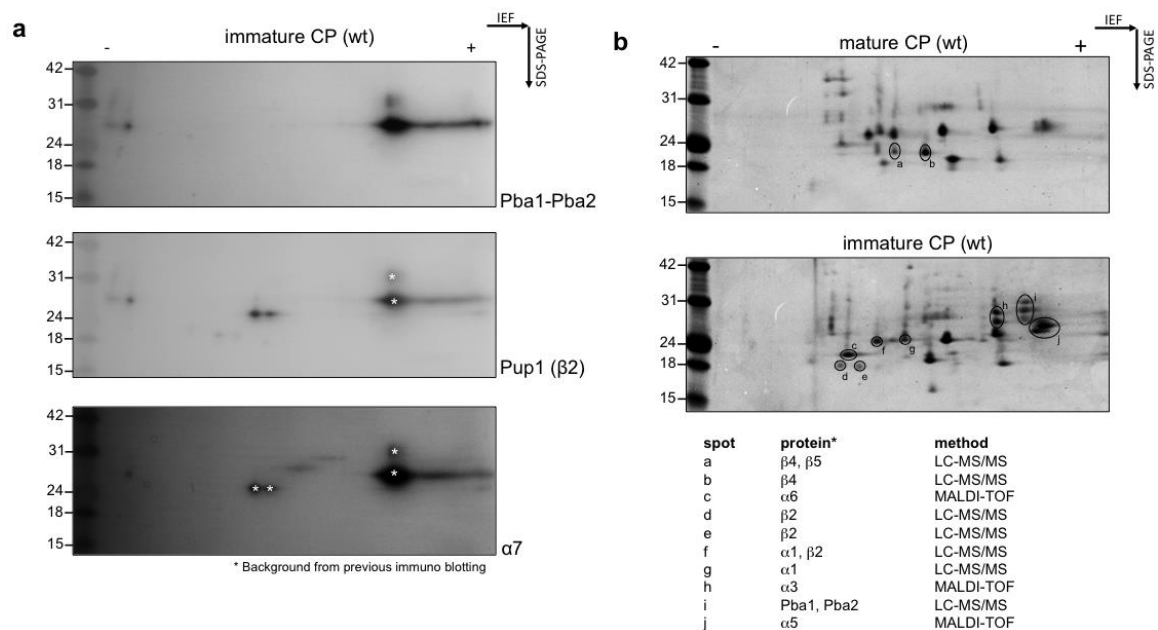
Supplementary Figure 1. Pba1-Pba2, but not Blm10, prevents RP association with immature CP.

(a) Proteasomes were purified from indicated strains using an affinity-tag on the $\beta 4$ subunit. Samples were resolved on SDS-PAGE and stained with CBB. In this preparation some Blm10 was cleaved at the N-terminus, causing the presence of a second faster migrating species of Blm10. **(b)** Pba1 and His-tagged Pba2 co-purified from *E.coli* were resolved on SDS-PAGE and immunoblotted using the serum (#1940) from a rabbit immunized with Pba1-Pba2 to show the antibody recognizes both Pba1 and Pba2. **(c)** Deletion of Blm10 does not cause an association of RP with immature CP. Immature CP from indicated strains was purified and resolved on SDS-PAGE followed by immunoblotting for indicated proteins. The enrichment in RP (Rpt1 blot) on immature CP observed for *pba1*Δ strains was not observed upon deletion of *BLM10*.



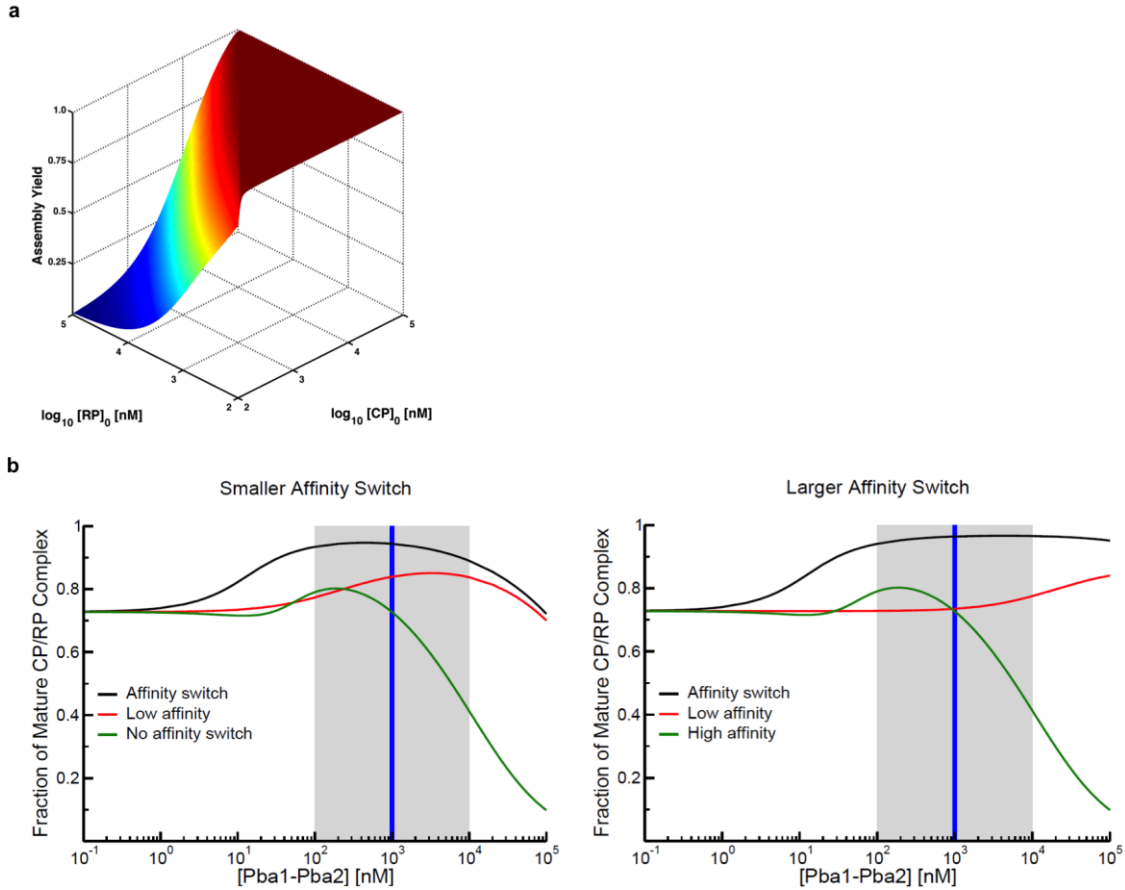
Supplementary Figure 2. Pba1-Pba2 and RP binding to immature and mature CP.

(a). *E. coli* expressed and purified His-tagged Pba1-Pba2 dimer was loaded onto a Ni²⁺-NTA sensor tip for the BLITZ (ForteBio). Next, association (175-300 s.) and dissociation (300- 440 s) of CP was monitored at increasing concentrations (starting at 0.22 nM and doubling up to 1.8 μ M). Analysis of the complete data set with baseline corrections yielded a K_D of 1.2 μ M ($K_a = 8.6 \pm 0.8 \times 10^4 \text{ M}^{-1}\text{s}^{-1}$ and $K_d 0.10 \pm 0.04 \text{ s}^{-1}$). **(b)** Titrations of Pba1-Pba2 to determine saturated binding on CP. CP from β 2-YFP tagged strains was purified using an affinity-tag present on the β 4 subunit. Next, purified CP was incubated with increasing amounts of His-tagged Pba1-Pba2 dimer for 30 minutes at 30 °C. Samples were resolved on native gel and stained in gel for suc-LLVY-AMC (left panel) or analyzed for YFP signal using a Typhoon 9410 imager (middle panel). Gels were also transferred to membranes and immunoblotted for the presence of Pba1-Pba2 (right panel). At 5 fold molar excess maximum CP binding was observed. **(c)** Mature and immature CP were analyzed on native gel for suc-LLVY-AMC hydrolytic activity (left panel). Gels were transferred to pvdF membrane and immunoblotted for indicated proteins. **(d)** Samples from **(c)** were used in a reconstitution assay with the base (an RP subcomplex) in 10 fold molar excess. Reconstitution assays were resolved by native gel electrophoresis followed by in gel suc-LLVY-AMC activity assay or immunoblotting (α 7 is a CP subunit and Rpt1 is a base subunit). Data show that the base reconstitutes efficiently on mature CP (lane 2), however, no reconstitution onto immature CP was observed (compare lane 2 with 4 and 5 in right panel).



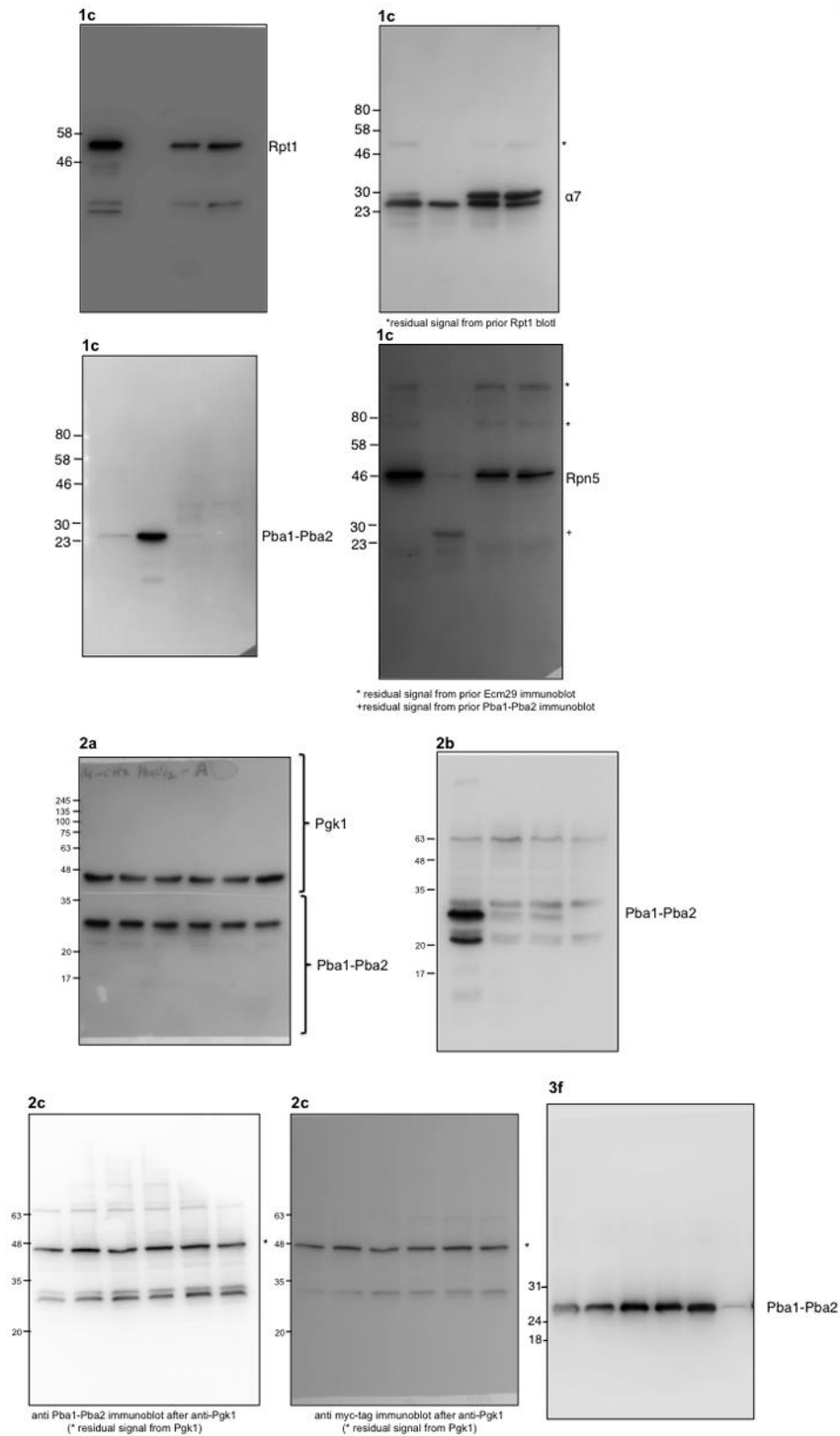
Supplementary Figure 3. Immunoblotting and mass spectrometry analysis of 2D gels from Figure 4 main text.

(a) 2D-gel electrophoresis of immature CP from a wildtype strain (see Fig. 4C main text) was subjected to immunoblotting. Panels show sequential probing of the membrane for Pba1-Pba2 (top panel), $\beta 2$ (middle panel) and $\alpha 7$ (lower panel). Spots indicated with an asterisk are signals derived from prior immunoblot, newly obtained signals are consistent with the expected PI and MW of proteins probed for. **(b)** Mass Spectrometry analysis of spots on 2D gel. Indicated spots were excised and submitted for analysis by MALDI-TOF, spots for which no result was obtained were subsequently analyzed by LC-MS/MS. MALDI-TOF analyses each time identified only one protein with significant score ($p < 0.05$). For LC-MS/MS only the identified proteins that contributed more than $>20\%$ to the total sample spectral count are reported to eliminate low abundant background contamination.



Supplementary Figure 4. Model of assembly is robust to variation and changes in parameters.

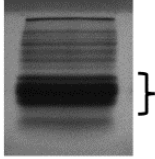
(a) Assembly yield is robust to variations in RP and CP concentration in the model. These results are from the mathematical model (see main text and other supplemental material). Aside from variations in total RP and CP concentration ($[RP]_0$ and $[CP]_0$, respectively), all the parameters are identical to those used for the "Affinity Switch" model in the main text. The concentration of the chaperone Pba1-Pba2 was held constant at 1 μ M. Since the CP and RP are at different concentrations in this case, the assembly yield is defined as the concentration of the RP-CP complex divided by the total concentration of RP or CP, depending on which is smaller. We find that near 100% yield is obtained for a wide variety of concentrations of both RP and CP, indicating that the results in Figure 5 of the main text do not depend on a specific set of RP and CP concentrations. **(b)** Changes in the magnitude of the affinity switch do not qualitatively effect our predictions. Left panel; similar to Figure 5a in the main text, but with an affinity switch that is 100 times instead of 1,000 times. In this model, the K_D of Pba1-Pba2 for the immature CP is unchanged at 1 nM, but it binds more tightly to the mature CP ($K_D = 100$ nM). Right panel; as before, now with an affinity switch that is 10,000 times. The K_D of the interaction with the immature CP is again unchanged at 1 nM, but the K_D of the mature interaction is weaker at 10 μ M. Note that in both cases, there is a broad range of Pba1-Pba2 concentrations that provides near 100% assembly yields. Modifying the affinity switch by making the K_D to the immature form stronger (or weaker) give similar results (data not shown).



Supplementary Figure 5. Uncropped images of the most important immunoblots from the main text.

Supplementary Table 1 Mass spectrometry analysis of immature CP purified from Ump1-TAP tagged strain.

Ump1-TAP
purification



Proteasome subunit	Systematic Gene Name	Standard name	# unique sequences	# total spectra
α 1	YGL011C	Scf1	16	28
α 2	YML092C	Pre8	8	11
α 3	YGR135W	Pre9	12	24
α 4	YOL038W	Pre6	10	15
α 5	YGR253C	Pup2	11	29
α 6	YMR314W	Pre5	9	19
α 7	YOR362C	Pre10	9	11
β 1	YJL001W	Pre3	5	5
β 2	YOR157C	Pup1	8	29
β 3	YER094C	Pup3	6	18
β 4	YER012W	Pre1	4	6
β 5	YPR103W	Pre2	8	10
β 6	YBL041W	Pre7	n.d.	n.d.
β 7	YFR050C	Pre4	n.d.	n.d.
Ump1	YBR173C	Ump1	7	8
Pba1	YLR199C	Pba1	13	29
Pba2	YKL206C	Add66	7	13

Sequences that cover the propeptides

β 2	M.AGLSFDNYQR.N R.NNFLAENSHTQPK.A
β 5	R.LAPSLTVPPIASPQQFLR.A K.ELQYDNEQNLESDFVTGASQFQR.L

Supplementary Table 2. Strains list.

Strain	*	Genotype	Figure	Ref.
sUB61	A	MAT α <i>lys2-801 leu2-3, 2-112 ura3-52 his3-Δ200 trp1-1</i>	2A-B	(b)
sDL135	A	MAT α <i>pre1::PRE1-TEVProA(HIS3)</i>	1A-D, 3A-B, 3E, 4C, S1A, S2A, S2C-D, S3B.	(c)
sDL133	A	MAT α <i>rpn11::RPN11-TEVProA(HIS3)</i>	3B.	(d)
SY36	A	MAT α <i>rpt1::HIS3, ProA-TEV-RPT1 in pEL36 (TRP)</i>	S2C-D.	(c)
Ump1Tap	B	MAT A <i>ump1::UMP1- CBP-TEV-ZZ-(His3MX6)</i>	1C-D, 3C, 4A, 4C, S1C, S2C- D, S3.	(e)
sJR789	A	MAT A <i>pba1::NAT pre1::PRE1-TEVProA(HIS3)</i>	S1A.	(a)
sJR790	A	MAT A <i>pba2::HYG pre1::PRE1-TEVProA(HIS3)</i>	S1A.	(a)
sJR601	A	MAT α <i>pba1::HYG pba2::NAT</i>	2B	(a)
sJR605	A	MAT A <i>pba1:: HYG pba2::NAT pre1::PRE1- TEVProA(HIS3)</i>	1A-B, S1A.	(a)
sJR791	A	MAT A <i>blm10::NAT rpn11::RPN11-TEVProA(HIS3)</i>	S1A.	(a)
sJR792	B	MAT A <i>pba1::NAT ump1::UMP1- CBP-TEV-ZZ- (His3MX6)</i>	1C, 4C, S1C.	(a)
sJR793	B	MAT A <i>pba1::HYG blm10::NAT ump1::UMP1- CBP- TEV-ZZ-(His3MX6)</i>	1C-D, 3D, S1C.	(a)
sJR794	B	MAT A <i>blm10::NAT ump1::UMP1- CBP-TEV-ZZ- (His3MX6)</i>	S1C.	(a)
sJR795	B	MAT A <i>pba1::PBA1ΔCT3 (KAN) ump1::UMP1- CBP- TEV-ZZ-(His3MX6)</i>	3F.	(a)
sJR782	B	MAT A <i>pba2::PBA2ΔCT3 (KAN) ump1::UMP1- CBP- TEV-ZZ-(His3MX6)</i>	3F.	(a)
sJR809	B	MAT A <i>pba1::PBA1ΔCT3 (KAN) pba2::PBA2ΔCT3 ump1::UMP1- CBP-TEV-ZZ-(His3MX6)</i>	3F.	(a)
sJR797	A	MAT α <i>pup1::PUP1-YFP (KAN) pre1::PRE1- TEVProA(HIS3)</i>	3B, S2B.	(a)
sJR852	A	MAT A <i>pba2::NAT</i>	2B	(a)
sJR853	A	MAT A <i>pba2::NAT + pCEN-His-Myc-Pba2(URA)</i>	2C	(a)
sJR857	A	MAT α <i>pba1::HYG</i>	2B	(a)

*All "A" strains have background genotype (*lys2-801 leu2-3, 2-112 ura3-52 his3- Δ 200 trp1-1*)

All "B" strains have BY4741 background genotype (MAT A *his3 Δ 0 leu2 Δ 0 met15 Δ 0 ura3 Δ 0*).

(a) This Study

(b) Finley, D., Ozkaynak, E. & Varshavsky, A. 1987 Cell 48, 1035-1046

(c) Leggett, D. S. et al. **2002** Mol Cell 10, 495-507

(d) Leggett, D. S., Glickman, M. H. & Finley, D. **2005** Methods Mol Biol 301, 57-70,

(e) Ghaemmaghami, S. et al. **2003** Nature 425, 737-741

Supplementary Note 1. Model for proteasome assembly

The model that we constructed for proteasome assembly draws heavily on the model we previously developed to describe the assembly of ring-like protein complexes¹. In this case we represented core particle (CP) assembly as the assembly of the α ring, which in eukaryotes is a heteromeric seven-member ring (e.g. $\alpha_1 - \alpha_7$). The assembly process in this case is derived from the following basic rules:

1. Assembly of the α ring is nucleated by the formation of an $\alpha_5:\alpha_6$ dimer from the respective monomers. In eukaryotic cells, this process is initiated by the binding of the dimeric chaperone Pba3-Pba4 to these two α subunits^{2,3}. For simplicity, we kept the role of Pba3-Pba4 *implicit*, representing its action by allowing the α_5 and α_6 monomers to spontaneously dimerize.
2. After formation of the $\alpha_5:\alpha_6$ dimer, either the α_4 or α_7 monomers could bind to the growing ring, generating the $\alpha_4:\alpha_5:\alpha_6$ or $\alpha_5:\alpha_6:\alpha_7$ trimers, respectively. In this model, all α subunits are monomeric until they associate with the "correct" end of the growing ring. All interactions are also considered to be perfectly specific: i.e. α_2 can only interact with α_1 and α_3 (its neighbors in the α ring), and not any other subunits³.
3. Completion of the entire α ring is considered an irreversible step. This is due in part to the fact that fully-formed rings are incredibly thermodynamically stable and thus do not tend to dissociate on physiologically relevant time scales^{1,4,5}. In CP assembly, completion of the α ring allows the β subunits to begin assembling on the ring; the Pba3-Pba4 chaperone dissociates from the complex after that process begins, and the chaperone Ump1 binds and assists with β ring formation and the eventual maturation of the CP^{2,3}. For simplicity we consider CP maturation to occur upon α ring completion.
4. The Pba1-Pba2 chaperone dimer can bind to any complex in the model that includes both α_5 and α_6 . Binding of this chaperone to any immature CP complex (i.e. any complex that is not a full α ring) occurs with some K_D that is considered distinct from the K_D of the interaction with the mature CP. This allows us to implement the affinity switch described in the main text.
5. For simplicity, we do not explicitly consider regulatory particle (RP) assembly in this model. All RP molecules are considered fully assembled and functional, since the RP assembly chaperones (e.g. Nas2) prevent binding of CP subunits to immature RP molecules⁶. Interaction of the RP with α monomers is considered to be fairly weak, while interactions with larger α ring complexes is considered to be strong. This latter consideration arises from the fact that the RP can make a large number of contacts with the surface of the α ring⁷, resulting in a fairly stable complex^{1,4,5}.
6. Binding of Pba1-Pba2 and the RP to any α complex is considered to be *mutually exclusive*. In other words, if an α ring subcomplex is bound to Pba1-Pba2, the RP cannot bind; similarly, if the RP is bound to a complex, Pba1-Pba2 cannot bind.

We used a procedure similar to the one we previously described to enumerate the set of chemical species allowed in this model, and the reactions that could occur between them, subject to the rules described above¹.

Supplementary Note 2. Ordinary Differential Equations

2.1 Notation

In the equations that follow, we use the "." character to denote a bound complex. So, for instance, $\alpha_5.\alpha_6$ is the α_5 - α_6 dimer, and $\alpha_5.\alpha_6.PBA_{1/2}$ is the same dimer bound to Pba1-Pba2. The following are the association rates in the model:

$k_{\alpha-\alpha}$: Association rate for α subunits binding to a growing α ring.

$k_{\alpha-\alpha.PBA_{1/2}}$: Association rate for α subunits binding to a growing α ring that is bound to a Pba1-Pba2 chaperone.

$k_{\alpha-\alpha.RP}$: Association rate for α subunits binding to a growing α ring that is bound to the RP.

$k_{PBA_{1/2}-\alpha}$: Association rate for Pba1-Pba2 binding to the α ring.

$k_{RP-\alpha}$: Association rate for the RP binding to the α ring.

All of these association rates are set to be equal in this model (following ref. ¹), and are given a value of $10^5 M^{-1}s^{-1}$. The dissociation rates in the model are:

$k_{\alpha-\alpha,n}$: Dissociation of α subunits from an incomplete α ring.

$k_{\alpha-\alpha.PBA_{1/2},n}$: Dissociation of α subunits from incomplete α rings that contain Pba1-Pba2.

$k_{\alpha-\alpha.RP,n}$: Dissociation of α subunits from incomplete rings that contain RP.

$k_{PBA_{1/2}-\alpha,n}$: Dissociation of Pba1-Pba2 from an incomplete α ring.

$k_{PBA_{1/2}-\alpha Ring,n}$: Dissociation of Pba1-Pba2 from a complete α ring.

$k_{RP-\alpha,n}$: Dissociation of the RP from either a complete or incomplete α ring.

The dissociation rates are set to obtain the K_0 's indicated in the text and figure legends. Note that we have formally included dissociation rates for RP-bound α subunits; as mentioned in the rules above, however, these rates are set to 0. This model explicitly considers synthesis of protein subunits and their loss from the system due to dilution; the degradation rate is k_{deg} , and is set so that the half-life of all proteins in the model is 2 hours, the approximate doubling time of yeast cells. Note that we do not consider transient changes in total protein concentration, so the total amount of each protein (α_1 , α_2 , $PBA_{1/2}$, ...) is fixed in time. To achieve this, the monomer is synthesized at a rate that *precisely* matches the rate of degradation of all of the complexes in which that particular subunit is found. For instance, the α_1 subunit is found in a set of complexes like α_1 (e.g. the monomer), $\alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}$, etc. All of these complexes are lost from the system due to dilution at the rate k_{deg} , and the α_1 monomer is synthesized at a rate that exactly cancels this dilution rate. As a result, all non-monomeric complexes have negative k_{deg} terms in their equations. All of the monomers also have positive terms in their equations, one for each dilution term that involves that monomer. This allows us to set some total protein concentration (say, $[Pba1-Pba2]_0$ in Figure 5 of the main text) that will remain invariant throughout the dynamics.

The ODEs describing the concentration of monomers are:

$$\begin{aligned} \frac{d\alpha_2}{dt} = & (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot k_{\alpha-\alpha,n} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7 \cdot k_{\alpha-\alpha,n} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{\alpha-\alpha,n} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{\alpha-\alpha,n}) \\ & + (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12,n}} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12,n}} \\ & + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12,n}} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12,n}}) \\ & + (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP \cdot k_{\alpha-\alpha.RP,n} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{\alpha-\alpha.RP,n} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{\alpha-\alpha.RP,n} \\ & + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{\alpha-\alpha.RP,n}) - (\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot \alpha_2 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha}) \\ & - (\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA_{12}} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA_{12}} \\ & + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA_{12}}) - (\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} + \alpha_1.\alpha_5.\alpha_6.\alpha_7.RP \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP}) \\ & + (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7 \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot k_{deg}) \\ & + (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{deg}) + (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP \cdot k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP \cdot k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP \cdot k_{deg}) \end{aligned}$$

$$\begin{aligned} \frac{d\alpha_6}{dt} = & (\alpha_5.\alpha_6.k_{\alpha-a,n}) - (\alpha_5.\alpha_6.k_{\alpha-\alpha}) + (\alpha_5.\alpha_6.k_{deg} + \alpha_5.\alpha_6.\alpha_7.k_{deg} \\ & + \alpha_1.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_4.\alpha_5.\alpha_6.k_{deg} \\ & + \alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.k_{deg} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.k_{deg}) + (\alpha_5.\alpha_6.PBA_{1/2}.k_{deg} + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} \\ & + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} \\ & + \alpha_4.\alpha_5.\alpha_6.PBA_{1/2}.k_{deg} + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}.k_{deg} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} \\ & + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}.k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}.k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}.k_{deg}) + (\alpha_5.\alpha_6.RP.k_{deg} + \alpha_5.\alpha_6.\alpha_7.RP.k_{deg} \\ & + \alpha_1.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_4.\alpha_5.\alpha_6.RP.k_{deg} \\ & + \alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.RP.k_{deg} \\ & + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP.k_{deg} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP.k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP.k_{deg})) \end{aligned}$$

We assume that α_5 and α_6 will form a trimer with $PBA_{3/4}$ in a non-rate limiting step. As such, we do not explicitly mention $PBA_{3/4}$ in the remaining ODEs, even though it would be bound to any complex with the α_5, α_6 dimer. The ODE for the α_5, α_6 complex is:

$$\begin{aligned} \frac{d\alpha_5, \alpha_6}{dt} = & (\alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} + \alpha_5 \cdot \alpha_6 \cdot \alpha_4 \cdot k_{\alpha-\alpha} \\ & + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} + \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} \\ & + \alpha_5 \cdot \alpha_6 \cdot RP \cdot k_{RP-\alpha} + \alpha_5 \cdot \alpha_6 \cdot k_{deg}) \end{aligned}$$

The ODEs describing the concentration of 3- α complexes are:

$$\begin{aligned} \frac{d\alpha_4, \alpha_5, \alpha_6}{dt} = & (\alpha_5 \cdot \alpha_6 \cdot \alpha_4 \cdot k_{\alpha-\alpha} + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{deg} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_3 \cdot k_{\alpha-\alpha} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot RP \cdot k_{RP-\alpha}) \\ \frac{d\alpha_5, \alpha_6, \alpha_7}{dt} = & (\alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{deg} + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_4 \cdot k_{\alpha-\alpha} + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_1 \cdot k_{\alpha-\alpha} \\ & + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot RP \cdot k_{RP-\alpha}) \end{aligned}$$

The ODEs describing the concentration of 4- α complexes are:

$$\begin{aligned} \frac{d\alpha_3, \alpha_4, \alpha_5, \alpha_6}{dt} = & (\alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_3 \cdot k_{\alpha-\alpha} + \alpha_2 \cdot \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{deg} + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_2 \cdot k_{\alpha-\alpha} + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} \\ & + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot RP \cdot k_{RP-\alpha}) \\ \frac{d\alpha_4, \alpha_5, \alpha_6, \alpha_7}{dt} = & (\alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} + \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_4 \cdot k_{\alpha-\alpha} \\ & + \alpha_3 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_1 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{deg} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_3 \cdot k_{\alpha-\alpha} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_1 \cdot k_{\alpha-\alpha} + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} \\ & + \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot RP \cdot k_{RP-\alpha}) \\ \frac{d\alpha_1, \alpha_5, \alpha_6, \alpha_7}{dt} = & (\alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_1 \cdot k_{\alpha-\alpha} + \alpha_1 \cdot \alpha_2 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_1 \cdot \alpha_4 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha, n}}) \\ & - (\alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{deg} + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha, n} \\ & + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha} + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot \alpha_4 \cdot k_{\alpha-\alpha} \\ & + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12-\alpha}} + \alpha_1 \cdot \alpha_5 \cdot \alpha_6 \cdot \alpha_7 \cdot RP \cdot k_{RP-\alpha}) \end{aligned}$$

The ODEs describing the concentration of 5- α complexes are:

[illegible]

The ODEs describing the concentration of 6- α complexes are:

[illegible]

The ODE describing the concentration of the full 7 member α ring is:

$$\begin{aligned} \frac{d\alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7}{dt} = & (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_1 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_2 \cdot k_{\alpha-\alpha} \\ & + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_3 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7 \cdot \alpha_4 \cdot k_{\alpha-\alpha} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot \alpha_7 \cdot k_{\alpha-\alpha} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12}-\alpha Ring,n}) \\ & - (\alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot k_{deg} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12}-\alpha Ring} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot RP \cdot k_{RP-\alpha}) \end{aligned}$$

The ODEs describing the concentration of the 2- α complex bound to $PBA_{1/2}$ is:

$$\begin{aligned} \frac{d\alpha_5.\alpha_6.PBA_{1/2}}{dt} = & (\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} \\ & + \alpha_5.\alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12}-\alpha}) \\ & - (\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA_{12}} \\ & + \alpha_5.\alpha_6.PBA_{1/2} \cdot k_{PBA_{12}-\alpha,n} + \alpha_5.\alpha_6.PBA_{1/2} \cdot k_{deg}) \end{aligned}$$

The ODEs describing the concentration of 3- α complexes bound to $PBA_{1/2}$ are:

$$\begin{aligned} \frac{d\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}}{dt} = & (\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} \\ & + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} + \alpha_4.\alpha_5.\alpha_6 \cdot PBA_{1/2} \cdot k_{PBA_{12}-\alpha}) \\ & - (\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} + \alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_3 \cdot k_{\alpha-\alpha.PBA_{12}} \\ & + \alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{PBA_{12}-\alpha,n} \\ & + \alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{deg}) \\ \frac{d\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}}{dt} = & (\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} \\ & + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} + \alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA_{12}-\alpha}) \\ & - (\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA_{12},n} + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA_{12}} \\ & + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_1 \cdot k_{\alpha-\alpha.PBA_{12}} + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{PBA_{12}-\alpha,n} \\ & + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg}) \end{aligned}$$

The ODEs describing the concentration of 4- α complexes bound to $PBA_{1/2}$ are:

$$\begin{aligned} \frac{d\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2}}{dt} &= (\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_3 \cdot k_{\alpha-\alpha.PBA12} + \alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} \\ &\quad + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_3.\alpha_4.\alpha_5.\alpha_6 \cdot PBA_{1/2} \cdot k_{PBA12-\alpha}) \\ &\quad - (\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA12} \\ &\quad + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA12} + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{PBA12-\alpha,n} \\ &\quad + \alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot k_{deg}) \\ \frac{d\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}}{dt} &= (\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA12} + \alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA12} \\ &\quad + \alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} \\ &\quad + \alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA12-\alpha}) \\ &\quad - (\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} \\ &\quad + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_3 \cdot k_{\alpha-\alpha.PBA12} + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_1 \cdot k_{\alpha-\alpha.PBA12} \\ &\quad + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{PBA12-\alpha,n} + \alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg}) \\ \frac{d\alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}}{dt} &= (\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_1 \cdot k_{\alpha-\alpha.PBA12} + \alpha_1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} \\ &\quad + \alpha_1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_1.\alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA12-\alpha}) \\ &\quad - (\alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{\alpha-\alpha.PBA12,n} + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA12} \\ &\quad + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA12} + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{PBA12-\alpha,n} \\ &\quad + \alpha_1.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg}) \end{aligned}$$

The ODEs describing the concentration of 5- α complexes bound to $PBA_{1/2}$ are:

[illegible]

The ODEs describing the concentration of 6- α complexes bound to $PBA_{1/2}$ are:

[illegible]

The ODE describing the concentration of the full 7 member α ring bound to $PBA_{1/2}$ is:

$$\begin{aligned} \frac{d\alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2}}{dt} = & (\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_1 \cdot k_{\alpha-\alpha.PBA12} + \alpha_1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_2 \cdot k_{\alpha-\alpha.PBA12} \\ & + \alpha_1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_3 \cdot k_{\alpha-\alpha.PBA12} + \alpha_1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot \alpha_4 \cdot k_{\alpha-\alpha.PBA12} \\ & + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.PBA_{1/2} \cdot \alpha_7 \cdot k_{\alpha-\alpha.PBA12} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7 \cdot PBA_{1/2} \cdot k_{PBA12-\alpha Ring}) \\ & - (\alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{PBA12-\alpha Ring,n} + \alpha_1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.PBA_{1/2} \cdot k_{deg}) \end{aligned}$$

The ODE describing the concentration of 2- α complexes bound to RP is:

$$\begin{aligned} \frac{d\alpha_5.\alpha_6.RP}{dt} = & (\alpha_4.\alpha_5.\alpha_6.RP \cdot k_{\alpha-\alpha.RP,n} + \alpha_5.\alpha_6.\alpha_7.RP \cdot k_{\alpha-\alpha.RP,n} \\ & + \alpha_5.\alpha_6 \cdot RP \cdot k_{RP-\alpha}) \\ & - (\alpha_5.\alpha_6.RP \cdot \alpha_4 \cdot k_{\alpha-\alpha.RP} + \alpha_5.\alpha_6.RP \cdot \alpha_7 \cdot k_{\alpha-\alpha.RP} \\ & + \alpha_5.\alpha_6.RP \cdot k_{deg}) \end{aligned}$$

The ODEs describing the concentration of 6- α complexes bound to RP are:

$$\begin{aligned}
\frac{d\alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP}}{dt} &= (\alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot \alpha_1 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - (\alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot k_{deg} + \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot k_{\alpha-\alpha.RP,n} \\
&\quad + \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot \alpha_7 \cdot k_{\alpha-\alpha.RP}) \\
\frac{d\alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP}}{dt} &= (\alpha_{3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} + \alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot \alpha_7 \cdot k_{\alpha-\alpha.RP} \\
&\quad + \alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - (\alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{deg} + \alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} \\
&\quad + \alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} + \alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_1 \cdot k_{\alpha-\alpha.RP}) \\
\frac{d\alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP}}{dt} &= (\alpha_{3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_1 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_3 \cdot k_{\alpha-\alpha.RP} \\
&\quad + \alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - (\alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{deg} + \alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} \\
&\quad + \alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} + \alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP}) \\
\frac{d\alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP}}{dt} &= (\alpha_{1.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_4 \cdot k_{\alpha-\alpha.RP} \\
&\quad + \alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - (\alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{deg} + \alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} \\
&\quad + \alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} + \alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_3 \cdot k_{\alpha-\alpha.RP}) \\
\frac{d\alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP}}{dt} &= (\alpha_{1.\alpha_2.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_3 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - (\alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{deg} + \alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{\alpha-\alpha.RP,n} \\
&\quad + \alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_4 \cdot k_{\alpha-\alpha.RP})
\end{aligned}$$

The ODE describing the concentration of the full 7 member α ring bound to RP is:

$$\begin{aligned}
\frac{d\alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP}}{dt} &= (\alpha_{2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_1 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_2 \cdot k_{\alpha-\alpha.RP} \\
&\quad + \alpha_{1.\alpha_2.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_3 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_2.\alpha_3.\alpha_5.\alpha_6.\alpha_7.RP} \cdot \alpha_4 \cdot k_{\alpha-\alpha.RP} \\
&\quad + \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.RP} \cdot \alpha_7 \cdot k_{\alpha-\alpha.RP} + \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6} \cdot RP \cdot k_{RP-\alpha}) \\
&\quad - \alpha_{1.\alpha_2.\alpha_3.\alpha_4.\alpha_5.\alpha_6.\alpha_7.RP} \cdot k_{deg}
\end{aligned}$$

Supplementary References

- 1 Deeds, E. J., Bachman, J. A. & Fontana, W. Optimizing ring assembly reveals the strength of weak interactions. *Proc Natl Acad Sci U S A* **109**, 2348-2353, (2012).
- 2 Murata, S., Yashiroda, H. & Tanaka, K. Molecular mechanisms of proteasome assembly. *Nat Rev Mol Cell Biol* **10**, 104-115 (2009).
- 3 Marques, A. J., Palanimurugan, R., Matias, A. C., Ramos, P. C. & Dohmen, R. J. Catalytic mechanism and assembly of the proteasome. *Chem Rev* **109**, 1509-1536 (2009).
- 4 Saiz, L. & Vilar, J. M. Stochastic dynamics of macromolecular-assembly networks. *Mol Syst Biol* **2**, 2006 0024 (2006).
- 5 Bray, D. & Lay, S. Computer-based analysis of the binding steps in protein complex formation. *Proc Natl Acad Sci U S A* **94**, 13493-13498 (1997).
- 6 Roelofs, J. *et al.* Chaperone-mediated pathway of proteasome regulatory particle assembly. *Nature* **459**, 861-865 (2009).
- 7 Beck, F. *et al.* Near-atomic resolution structural model of the yeast 26S proteasome. *Proc Natl Acad Sci U S A* **109**, 14870-14875 (2012).